

Neutrokin- α

1 AAATTCAGGATAACTCTCCTGAGGGGTGAGCCAAGCCCTGCCATGTAGTGCACGCAGGAC 60

61 ATCAACAAACACAGATAACAGGAAATGATCCATTCCCTGTGGTCACTTATTCTAAAGGCC 120

121 CCAACCTTCAAAGTTCAAGTAGTGATATGGATGACTCCACAGAAAGGGAGCAGTCACGCC 180
1 M D D S T E R E Q S R L 12

181 TTA CT TCT TGC CTTAAGAAAAGAGAAGAAATGAACTGAAGGAGTGTGTTTCCATCCTCC 240
13 T S C L K K R E E M K L K E C V S I L P 32
CD-I

241 CACGGAAGGAAAGCCCCTCTGTCCGATCCTCCAAAGACGGAAAGCTGCTGGCTGCAACCT 300
33 R K E S P S V R S S K D G K L L A A T L 52
CD-I

301 TGCTGCTGGCACTGCTGTCTTGCTGCCTCACGGTGGTGTCTTTCTACCAGGTGGCCGCC 360
53 L L A L L S C C L T V V S F Y Q V A A L 72

361 TGCAAGGGGACCTGGCCAGCCTCCGGGCAGAGCTGCAGGGCCACCACGCGGAGAAGCTGC 420
73 Q G D L A S L R A E L Q G H H A E K L P 92
CD-II

421 CAGCAGGAGCAGGAGCCCCAAGGCCGGCCTGGAGGAAGCTCCAGCTGTCAACGCGGGAC 480
93 A G A G A P K A G L E E A P A V T A G L 112
CD-III

481 TGAAAATCTTTGAACCACCAGCTCCAGGAGAAGGCAACTCCAGTCAGAACAGCAGAAATA 540
113 K I F E P P A P G E G N S S Q N S R N K 132
#

541 AGCGTGCCGTTCAAGGTCCAGAAGAAACAGTCACTCAAGACTGCTTGCAACTGATTGCAG 600
133 R A V Q G P E E T V T Q D C L Q L I A D 152
CD-IV

FIG.1A

Neutrokin- α

601 ACAGTGAAACACCAACTATACAAAAAGGATCTTACACATTTGTTCCATGGCTTCTCAGCT 660
 153 S E T P T I Q K G S Y T F V P W L L S F 172
 CD-V

661 TTAAAAGGGGAAGTGCCCTAGAAGAAAAAGAGAATAAAATATTGGTCAAAGAAACTGGTT 720
 173 K R G S A L E E K E N K I L V K E T G Y 192
 CD-V CD-VI

721 ACTTTTTTATATATGGTCAGGTTTTATATACTGATAAGACCTACGCCATGGGACATCTAA 780
 193 F F I Y G Q V L Y T D K T Y A M G H L I 212
 CD-VI CD-VII

781 TTCAGAGGAAGAAGGTCCATGTCTTTGGGGATGAATTGAGTCTGGTGACTTTGTTTCGAT 840
 213 Q R K K V H V F G D E L S L V T L F R C 232
 CD-VII CD-VIII

 841 GTATTCAAATATGCCTGAAACACTACCCAATAATTCCTGCTATTCAGCTGGCATTGCAA 900
 233 I Q N M P E T L P N N S C Y S A G I A K 252
 CD-VIII CD-IX

901 AACTGGAAGAAGGAGATGAACTCCAACCTTGCAATACCAAGAGAAAATGCACAAATATCAC 960
 253 L E E G D E L Q L A I P R E N A Q I S L 272
 CD-X

961 TGGATGGAGATGTCACATTTTTTGGTGCATTGAAACTGCTGTGACCTACTTACACCATGT 1020
 273 D G D V T F F G A L K L L 285
 CD-XI

1021 CTGTAGCTATTTTCCTCCCTTCTCTGTACCTCTAAGAAGAAAGAATCTAACTGAAAATA 1080

1081 CCAAAAAAAAAAAAAAAAAA 1100

FIG.1B

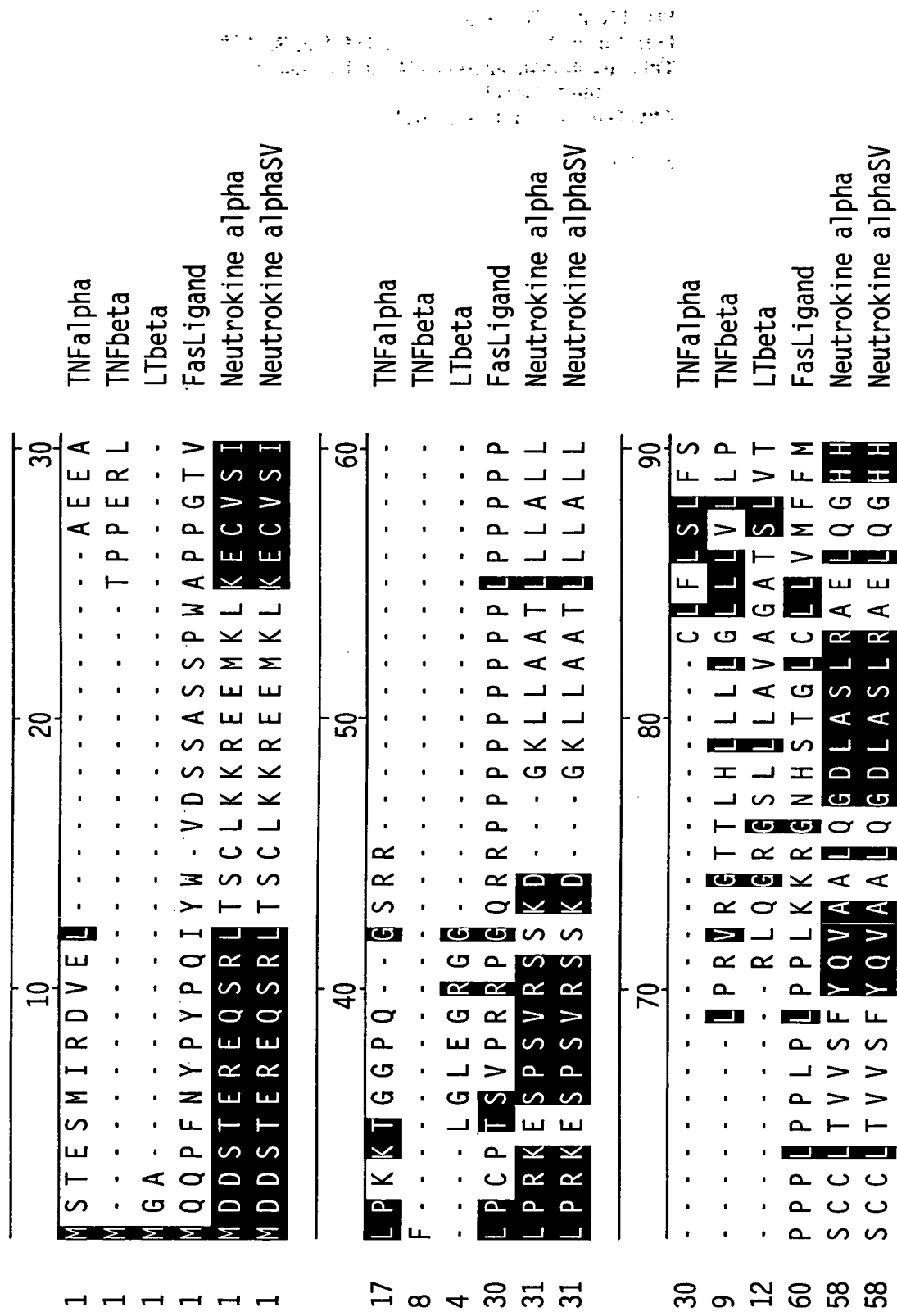


FIG.2A

	100	110	120	
38	FL - - I V A G A T T L F C L L H F G V I G P Q R E E F P R			TNFalpha
31	GA Q G L P G V G L - - - - - - - - - - - - - - - - -			TNFBeta
32	LL A V P I T V L A V L A L V P Q D Q G G L V T E T A D P			LTbeta
90	V L V A L V G L G L G M F Q L F H L Q K E L A E L R E S T S			FasLigand
88	A E K L P A G A G A P K A G L E E A P A V T A G L K I F F E P			Neutrokine alpha
88	A E K L P A G A G A P K A G L E E A P A V T A G L K I F F E P			Neutrokine alphaSV
	130	140	150	
66	D L S L I S - P L A - Q A V R S S S R T P S D - - - K P V A			TNFalpha
41	- - - T P S - A A Q - T A R Q H P K M H L A H S T L K P A A			TNFBeta
62	G A Q A Q Q - G L G F Q K L P E E E P E T D L S P G L P A A			LTbeta
120	Q M H T A S - S L E - K Q I G H P S P P P E K K E L R K V A			FasLigand
118	P A P G E G N S S Q N S R N K R A V Q G P E E T V T Q D C L			Neutrokine alpha
118	P A P G E G N S S Q N S R N K R A V Q G P E E T - - - - -			Neutrokine alphaSV
	160	170	180	
91	H V V A N P Q A E G - Q - - - - - L Q W L N R R A N A L L			TNFalpha
66	H L I G D P S K Q N - S - - - - - L L W R A N T D R A F L			TNFBeta
91	H L I G A P L K - G Q G - - - - - L G W E T T K E Q A F L			LTbeta
148	H L T G K S N S R S M P - - - - - L E W E D T Y G I V L L			FasLigand
148	Q L I A D S E T P T I Q K G S Y T F V P W L - - - L S F K			Neutrokine alpha
142	- - - - - - - - - - - G S Y T F V P W L - - - L S F K			Neutrokine alphaSV

114	A	N	G	V	E	L	R	D	N	-	Q	L	V	V	P	S	E	G	L	Y	L	I	Y	S	Q	V	L	F	K	G
89	Q	D	G	F	S	L	S	N	N	-	S	L	L	V	P	T	S	G	I	Y	F	V	Y	S	Q	V	V	F	S	G
114	T	S	G	T	Q	F	S	D	A	E	G	L	A	L	P	Q	D	G	L	Y	L	Y	C	L	V	G	Y	R	G	G
172	-	S	G	V	K	Y	K	K	G	-	G	L	V	I	N	E	T	G	L	Y	F	V	Y	S	K	V	Y	F	R	G
174	R	G	S	A	L	E	E	K	E	N	K	I	L	V	K	E	T	G	Y	F	F	I	Y	G	Q	V	L	Y	T	D
155	R	G	S	A	L	E	E	K	E	N	K	I	L	V	K	E	T	G	Y	F	F	I	Y	G	Q	V	L	Y	T	D

TNFa1pha
 TNFbeta
 LTbeta
 FasLigand
 Neutrokinine alpha
 Neutrokinine alphaSV

143	Q	G	C	P	-	-	-	-	-	-	S	T	H	V	L	L	T	H	T	I	S	R	I	A	V	S	Y	Q	T	K
118	K	A	Y	S	P	-	-	K	A	T	S	S	P	L	Y	L	A	H	E	V	Q	L	F	S	S	Q	Y	P	F	H
144	R	A	P	P	G	G	G	D	P	Q	G	R	S	V	T	L	R	S	S	L	Y	R	A	G	G	A	Y	G	P	G
200	Q	S	C	N	-	-	-	-	-	-	-	-	N	L	P	L	S	H	K	V	Y	M	R	N	S	K	Y	P	Q	D
204	K	T	Y	A	M	G	-	-	-	-	-	-	H	L	I	Q	R	K	K	V	H	V	F	G	D	E	L	S	-	-
185	K	T	Y	A	M	G	-	-	-	-	-	-	H	L	I	Q	R	K	K	V	H	V	F	G	D	E	L	S	-	-

TNFa1pha
 TNFbeta
 LTbeta
 FasLigand
 Neutrokinine alpha
 Neutrokinine alphaSV

167	V	N	-	-	L	L	S	A	I	K	S	P	C	Q	R	E	T	P	E	-	-	G	A	E	A	K	P	W	Y	E
146	V	P	-	-	L	L	S	S	Q	K	M	V	Y	P	-	-	-	-	-	-	-	G	L	Q	E	P	W	L	H	
174	T	P	E	L	L	E	G	A	E	T	V	T	P	V	L	D	P	A	R	R	Q	G	Y	G	P	L	W	Y	T	
222	L	V	-	-	M	M	E	G	K	M	M	S	Y	C	-	-	-	-	-	-	-	T	T	G	Q	M	W	A	R	
226	L	V	T	L	F	R	C	I	Q	N	M	P	E	T	L	P	N	-	-	-	-	-	-	-	-	-	-	-	N	
207	L	V	T	L	F	R	C	I	Q	N	M	P	E	T	L	P	N	-	-	-	-	-	-	-	-	-	-	-	N	

TNFa1pha
 TNFbeta
 LTbeta
 FasLigand
 Neutrokinine alpha
 Neutrokinine alphaSV

FIG.2C

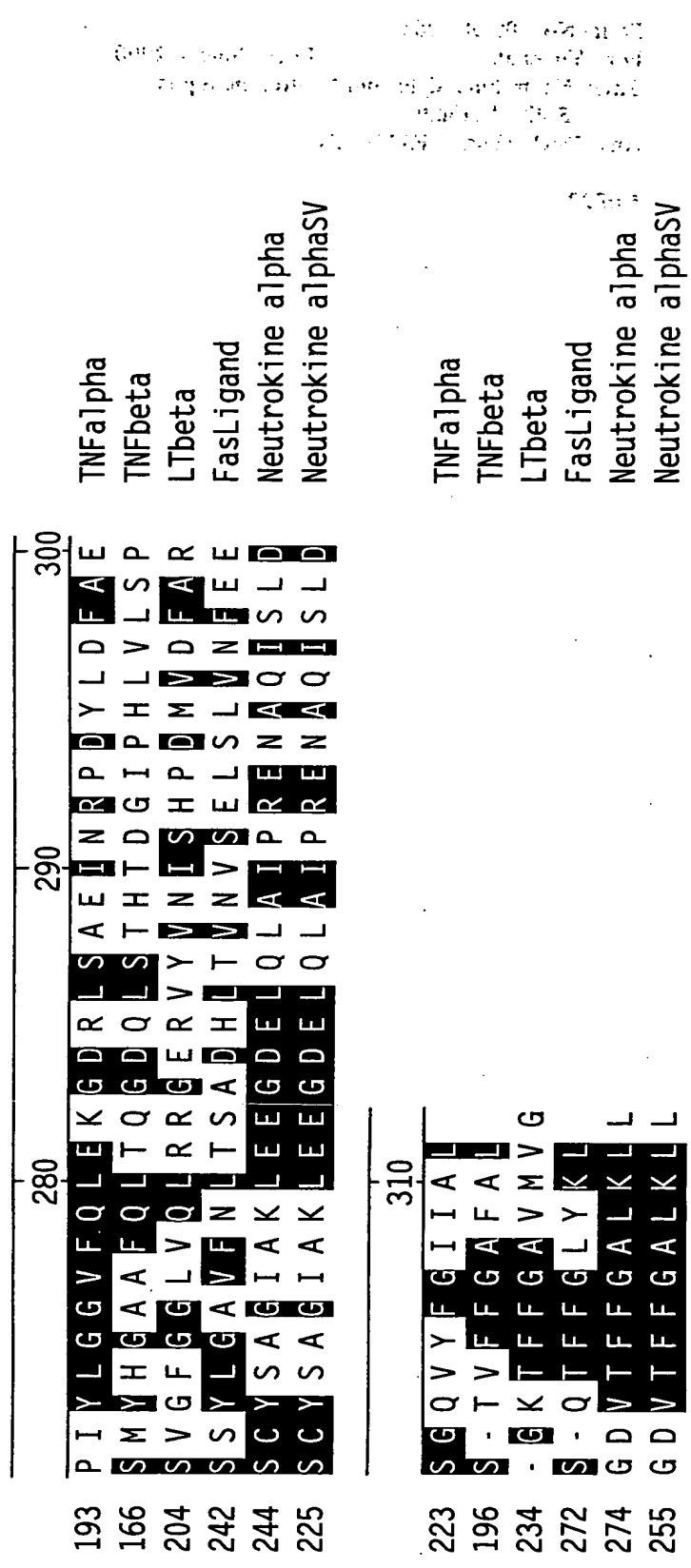
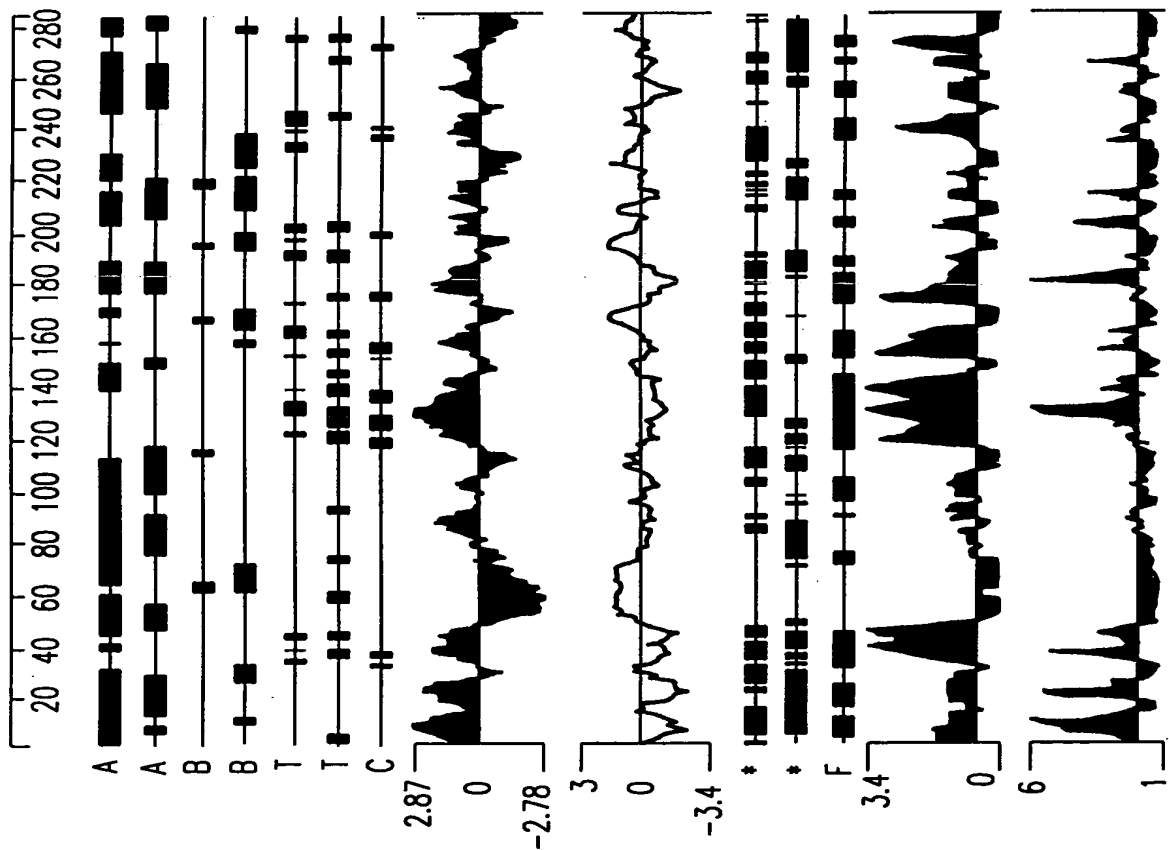


FIG.2D

NEUTROKINE- α



■ ALPHA, REGIONS-GARNIER-ROBSON

■ ALPHA, REGIONS-CHOU-FASMAN

■ BETA, REGIONS-GARNIER-ROBSON

■ BETA, REGIONS-CHOU-FASMAN

■ TURN, REGIONS-GARNIER-ROBSON

■ TURN, REGIONS-CHOU-FASMAN

■ COIL, REGIONS-GARNIER-ROBSON

■ HYDROPHILICITY PLOT-KYTE-DOOLITTLE

□ HYDROPHOBICITY PLOT-HOPP-WOODS

■ ALPHA, AMPHIPATHIC REGIONS-EISENBERG

■ BETA, AMPHIPATHIC REGIONS-EISENBERG

■ FLEXIBLE REGIONS-KARPLUS-SCHULZ

■ ANTIGENIC INDEX-JAMESON-WOLF

■ SURFACE PROBABILITY PLOT-EMINI

FIG.3

	1				50
HSOAD55RA	GGNTAACTCT	CCTGAGGGGT	GAGCCAAGCC	CTGCCATGTA
HNEDU15X	...AAATTCA	GGATAACTCT	CCTGAGGGGT	GAGCCAAGCC	CTGCCATGTA
HSLAH84R	.AATTCGGCA	NAGNAACTG	GTTACTTTT	TATATATGGT	CAGGTTTTAT
HLTBM08R	AATTCGGCAC	GAGCAAGGCC	GGCCTGGAGG	AAGCTCCAGC	TGTCACCGCG
	51				100
HSOAD55R	GTGCACGCAG	GACATCANCA	A..ACACANN	NNNCAGGAAA	TAATCCATTCT
HNEDU15X	GTGCACGCAG	GACATCAACA	A..ACACAGA	TAACAGGAAA	TGATCCATTCT
HSLAH84R	ATACTGATAA	GACCTACGCC	ATGGGACATC	TAGTTCAGAG	GAAGAAGGTC
HLTBM08R	GGACTGAAAA	TCTTTGAACC	ACCAGCTCCA	GGAGAAGGCA	ACTCCAGTCA
	101				150
HSOAD55R	CCTGTGGTCA	CTTATTCTAA	AGGCCCAAC	CTTCAAAGTT	CAAGTAGTGA
HNEDU15X	CCTGTGGTCA	CTTATTCTAA	AGGCCCAAC	CTTCAAAGTT	CAAGTAGTGA
HSLAH84R	CATGTCTTTG	GGGATGAATT	GAGTCTGGTG	ACTTTGTTTC	GATGTATTCA
HLTBM08R	GAACAGCAGA	AATAAGCGTG	CCGTTTCAGG	TCCAGAAGAA	ACAGTCACTC
	151				200
HSOAD55R	TATGGATGAC	TCCACAGAAA	GGGAGCAGTC	ACGCCTTACT	TCTTGCCCTTA
HNEDU15X	TATGGATGAC	TCCACAGAAA	GGGAGCAGTC	ACGCCTTACT	TCTTGCCCTTA
HSLAH84R	AAATATGCCT	GAAACACTAC	CCAATAATTC	CTGCTATTCA	GCTGGCATTG
HLTBM08R	AAGACTGCTT	GCAACTGNNT	GCAGACAGTG	AAACACCAAC	TATACAAAAA
	201				250
HSOAD55R	AGAAAAGAGA	AGAAATGAAA	CTGNAAGGAG	TGTGTTTCCA	TCCTCCCACG
HNEDU15X	AGAAAAGAGA	AGAAATGAAA	CT.GAAGGAG	TGTGTTTCCA	TCCTCCCACG
HSLAH84R	CAAACTGGN	AGGAAGGA..	...GATGAAC	TCCAAC TTGC	AATACCAGGG
HLTBM08R	GGCTCCCTTC	TGNTGCCACA	TTTGGGCCAA	GGAATGGAGA	GATTTCTTCG
	251				300
HSOAD55R	GAAGGAAAGC	CCCTCTNTCC	GATCCTCCAA	AGACGGAAAG	CTGCTGGCTG
HNEDU15X	GAAGGAAAGC	CCCTCTGTCC	GATCCTCCAA	AGACGGAAAG	CTGCTGGCTG
HSLAH84R	GAAAATGCAC	AATTATCACT	GGGATGGAGA	TGTTACATT	TTTTGGGTGC
HLTBM08R	TCTGGAAACA	TTTTGCCAAA	CTCTTCAGAT	ACTCTTNTCT	CTCTGGGAAT
	301				350
HSOAD55R	CAACCTTGNT	GNTGGCATTG	TGTTCTTGCT	GNCTCAAGGT	GGTGTNTT.
HNEDU15X	CAACCTTGCT	GCTGGCACTG	CTGTCTTGCT	GCCTCACGGT	GGTGTCTTTC
HSLAH84R	CATTGAACT	GCTGTGACCT	NCTTACANCA	NGTGCTGTTN	GCTATTTTNC
HLTBM08R	CAAAGGAAAA	TCTCTACTTA	GATTNACACA	TTTGTTCCCA	TGGGTNTCTT
	351				400
HSOAD55R
HNEDU15X	TACCAGGTGG	CCGCCCTGCA	AGGGGACCTG	GCCAGCCTCC	GGGCAGAGCT
HSLAH84R	CTNCCTNTTC	TNTGGTAACC	TCTTAGGAAG	GAAGGATTCT	TAAC TGGGAA
HLTBM08R	AAGTTTTAAA	AGGGGAGTGC	CCTTAGGAGG	AAAAGGGGAT	AAATATTGGC

FIG.4A

	801		850
HSOAD55R
HNEDU15X	GTCCATGTCT	TTGGGGATGA	ATTGAGTCTG GTGACTTTGT TTCGATGTAT
HSLAH84R
HLTBM08R
	851		900
HSOAD55R
HNEDU15X	TCAAAATATG	CCTGAAACAC	TACCCAATAA TTCCTGCTAT TCAGCTGGCA
HSLAH84R
HLTBM08R
	901		950
HSOAD55R
HNEDU15X	TTGCAAAACT	GGAAGAAGGA	GATGAACTCC AACTTGCAAT ACCAAGAGAA
HSLAH84R
HLTBM08R
	951		1000
HSOAD55R
HNEDU15X	AATGCACAAA	TATCACTGGA	TGGAGATGTC ACATTTTTTG GTGCATTGAA
HSLAH84R
HLTBM08R
	1001		1050
HSOAD55R
HNEDU15X	ACTGCTGTGA	CCTACTTACA	CCATGTCTGT AGCTATTTTC CTCCCTTTCT
HSLAH84R
HLTBM08R
	1051		1100
HSOAD55R
HNEDU15X	CTGTACCTCT	AAGAAGAAAG	AATCTAACTG AAAATACCAA AAAAAAAAAA
HSLAH84R
HLTBM08R
	1101		
HSOAD55R		
HNEDU15X	AAAAAA		
HSLAH84R		
HLTBM08R		

FIG.4C

Neutrokin-αSV

1	ATGGATGACTCCACAGAAAGGGAGCAGTCACGCCTTACTTCTTGCCTTAAGAAAAGAGAA	60
1	M D D S T E R E Q S R L T S C L K K R E	20
61	GAAATGAACTGAAGGAGTGTGTTTCCATCCTCCACGGAAGGAAAGCCCCTCTGTCCGA	120
21	E M K L K E C V S I <u>L P R K E S P S V R</u>	40
	CD-I	
121	TCCTCCAAAGACGGAAAGCTGCTGGCTGCAACCTTGCTGCTGGCACTGCTGTCTTGCTGC	180
41	<u>S S K D</u> G K <u>L L A A T L L L A L L S C C</u>	60
	CD-I	
181	CTCACGGTGGTGTCTTTCTACCAAGTGGCCGCCCTGCAAGGGGACCTGGCCAGCCTCCGG	240
61	<u>L T V V S F Y Q V A A L</u> Q G D L A S L R	80
	CD-II	
241	GCAGAGCTGCAGGGCCACCACGCGGAGAAGCTGCCAGCAGGAGCAGGAGCCCCAAGGCC	300
81	<u>A E L</u> Q G H H A E K L P A <u>G A G A P K A</u>	100
	CD-II CD-III	
301	GGCCTGGAGGAAGCTCCAGCTGTCACCGCGGGACTGAAAATCTTTGAACCACCAGCTCCA	360
101	<u>G L</u> E E A P A V T A G L K I F E P P A P	120
	CD-III	
	#	
361	GGAGAAGGCAACTCCAGTCAGAACAGCAGAAATAAGCGTGCCGTTCCAGGTCCAGAAGAA	420
121	G E G N S S Q N S R N K R A V Q G P E E	140
421	ACAGGATCTTACACATTTGTTCCATGGCTTCTCAGCTTTAAAAGGGGAAGTGCCCTAGAA	480
141	T G S Y T F <u>V P W L L S F K R G S A L E</u>	160
	CD-IV	
481	GAAAAAGAGAATAAAATATTGGTCAAAGAACTGGTTACTTTTTATATATGGTCAGGTT	540
161	<u>E K</u> E N K <u>I L V K E T G Y F F I Y G Q V</u>	180
	CD-IV CD-V	
541	TTATATACTGATAAGACCTACGCCATGGGACATCTAATTCAGAGGAAGAAGGTCCATGTC	600
181	<u>L Y T D K T Y A M G</u> <u>H L I Q R K K V H V</u>	200
	CD-VI CD-VII	

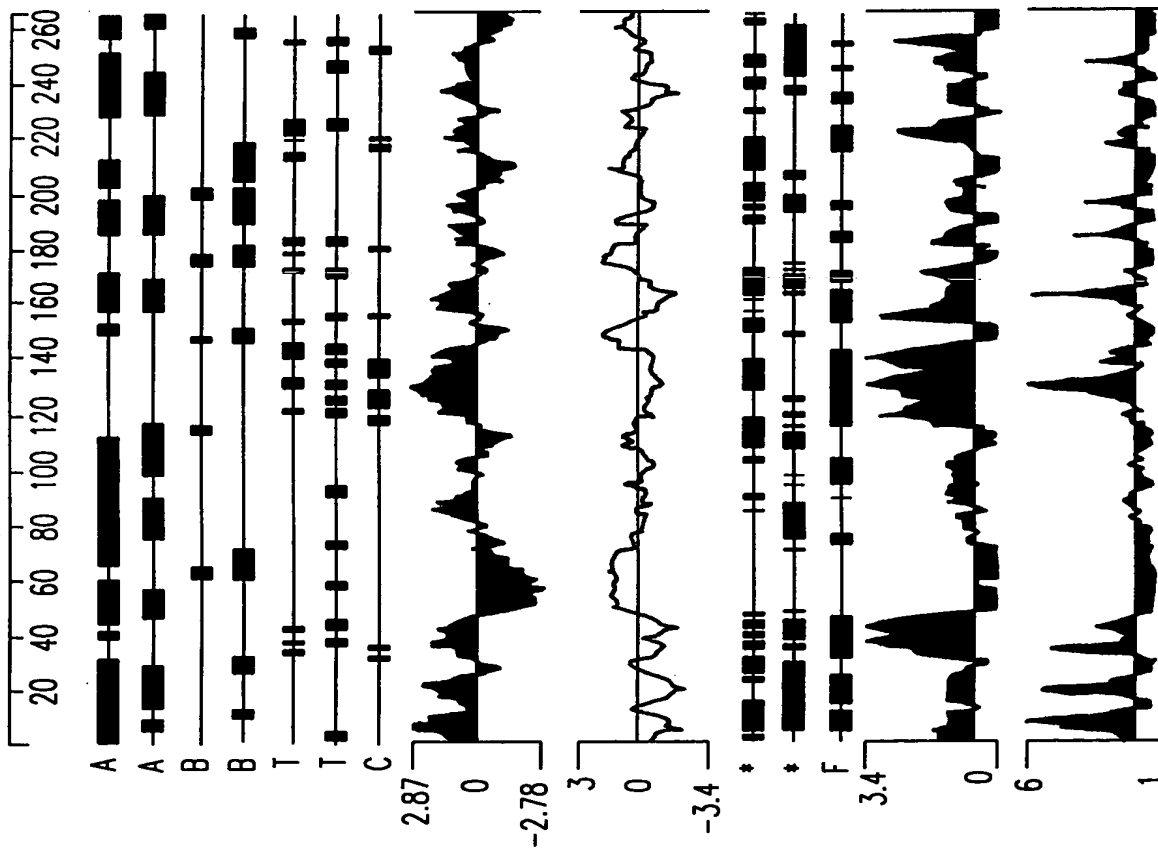
FIG.5A

Neutrokin- α SV

601 TTTGGGGATGAATTGAGTCTGGTGACTTTGTTTCGATGTATTCAAAATATGCCTGAAACA 660
 201 F G D E L S L V T L F R C I Q N M P E T 220
 CD-VIII CD-VIII
 661 CTACCCAATAATTCCTGCTATTCAGCTGGCATTGCAAACTGGAAGAAGGAGATGAACTC 720
 221 L P N N S C Y S A G I A K L E E G D E L 240
 CD-IX CD-X
 721 CAACTTGCAATACCAAGAGAAAATGCACAAATATCACTGGATGGAGATGTCACATTTTTT 780
 241 Q L A I P R E N A Q I S L D G D V T F F 260
 CD-X CD-XI
 781 GGTGCATTGAACTGCTGTGACCTACTTACACCATGTCTGTAGCTATTTTCCTCCCTTTC 840
 261 G A L K L L 266
 CD-XI
 841 TCTGTACCTCTAAGAAGAAAGAATCTAACTGAAAATACCAAAAAAAAAAAAAAAAAAAAA 900
 901 AAA 903

FIG.5B

NEUTROKINE- α SV



■ ALPHA, REGIONS-GARNIER-ROBSON
 ■ ALPHA, REGIONS-CHOU-FASMAN
 ■ BETA, REGIONS-GARNIER-ROBSON
 ■ BETA, REGIONS-CHOU-FASMAN
 ■ TURN, REGIONS-GARNIER-ROBSON
 ■ TURN, REGIONS-CHOU-FASMAN
 ■ COIL, REGIONS-GARNIER-ROBSON

■ HYDROPHILICITY PLOT-KYTE-DOOLITTLE

□ HYDROPHOBICITY PLOT-HOPP-WOODS

■ ALPHA, AMPHIPATHIC REGIONS-EISENBERG
 ■ BETA, AMPHIPATHIC REGIONS-EISENBERG
 ■ FLEXIBLE REGIONS-KARPLUS-SCHULZ

■ ANTIGENIC INDEX-JAMESON-WOLF

■ SURFACE PROBABILITY PLOT-EMINI

FIG.6

Neutrokin-

Alpha MDDSTEREQSRLTSCCLKREEMK LKECVSILPRKESPSVRS 41

Transmembrane Region

SKDGKLLAATLLALLSCCLTVVSVFYQVAALQGD LASLRAE 82

LQGHHA EKLPAGAGAPKAGLEEAPAVTAGLKIFEP P APGEG 123

↓
NSSQNSRNKRAVQGP EETVT QDCLQLIADSE TPTIQKGSYT 164
April HSVLHLLVPINATSK-DDSDVT 134
TNF KPVAAHVVANPQAEQQ----- 102
LT α KPAAHLIGDPSKQNS----- 76

A' B' B C
FV P W L L S - - - - F K R G S A L E E K E N K I L V K E T G Y F F I Y G Q V L 200
E V M W Q P A - - - - L R R G R G L Q A Q G Y G V R I Q D A G V Y L L Y S Q V L 170
- L Q W L N R R A N A L L A N G V E L R D - - N Q L V V P S E G L Y L I Y S Q V L 139
- L L W R A N T D R A F L Q D G F S L S N - - N S L L V P T S G I Y F V Y S Q V V 114

D E
Y T D K T Y - - - - A M G H L I Q R K K V H V F G D E L S L V T L F R C I Q N M P 237
F Q D V T F - - - - I M G Q V V S R E - - - - G Q G R Q E T L F R C I R S M P 201
F K G Q G C P - - - - S T H V L L T H I S R I A V S Y Q T K V N L L S A I K S P 176
F S G K A Y S P K A I S S P I Y L A H E V Q L F S S Q Y P F H V P L L S S Q K M V 155

FIG.7A-1

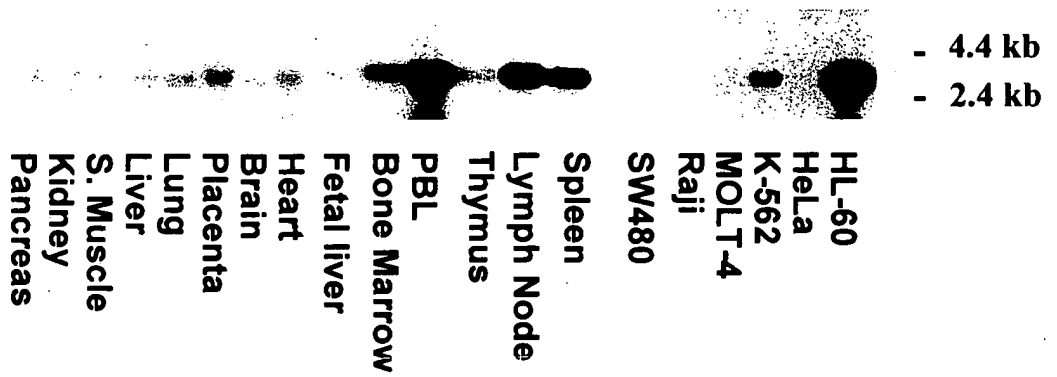


FIG.7B

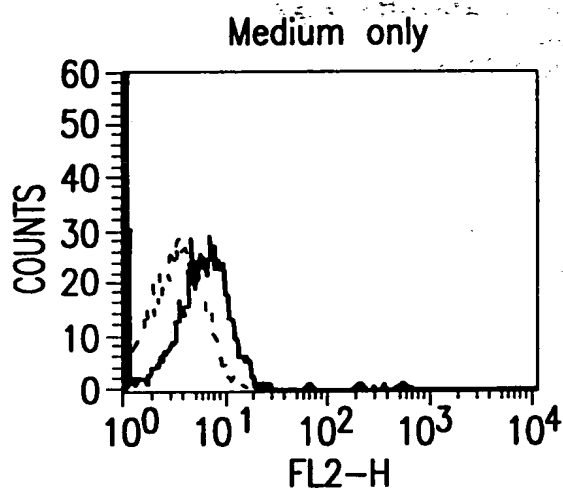


FIG.8A

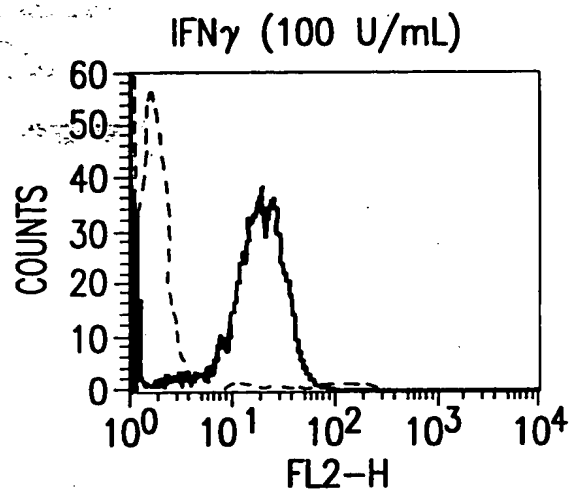


FIG.8B

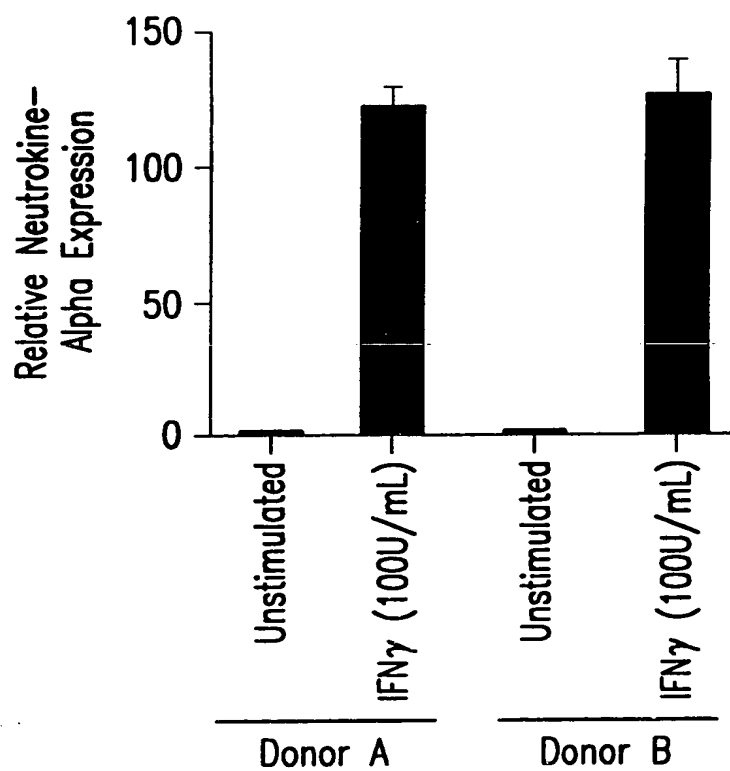


FIG.8C

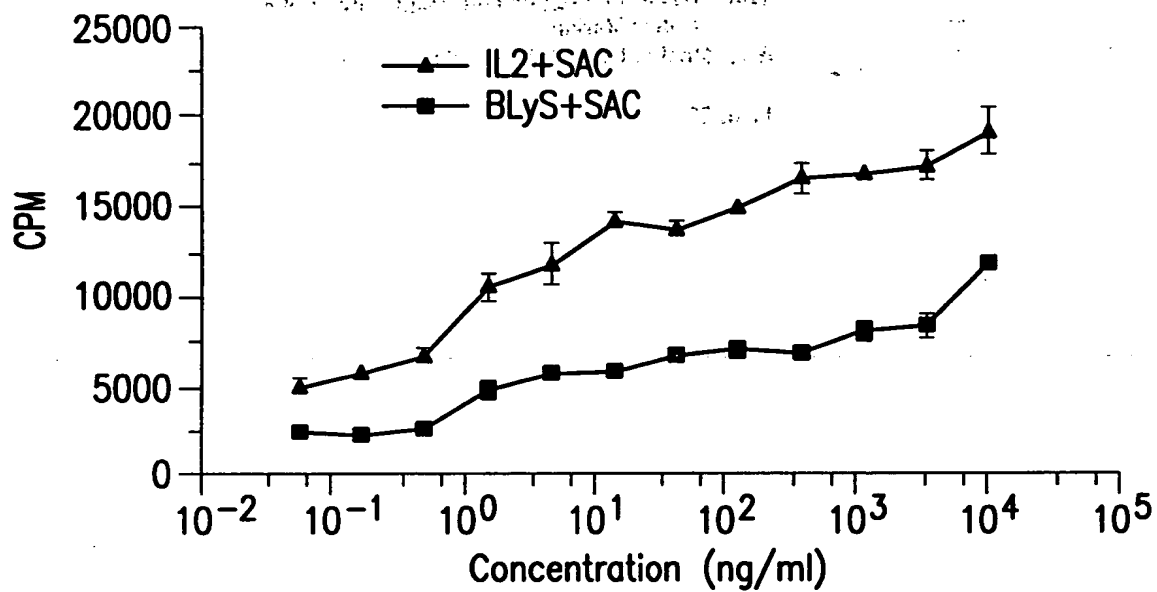


FIG. 9A

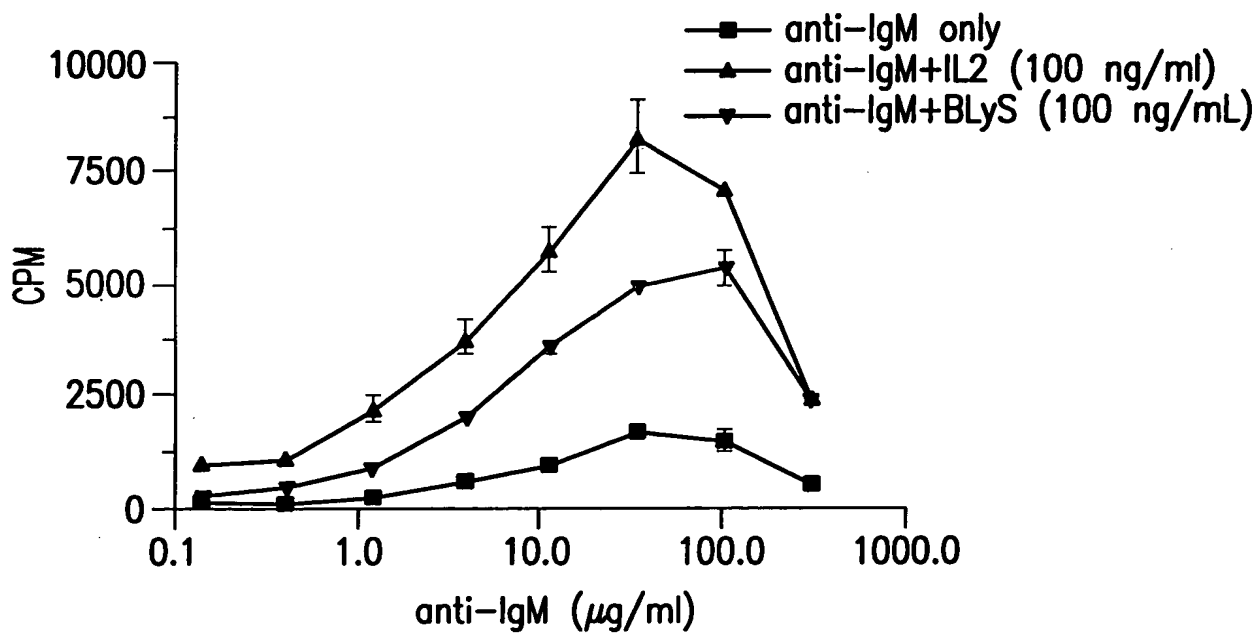


FIG. 9B

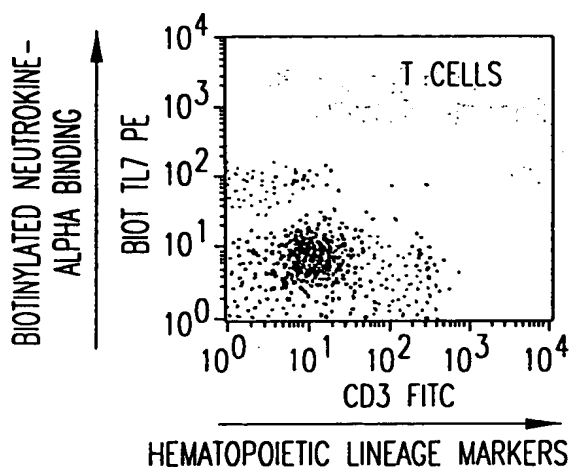


FIG.10A

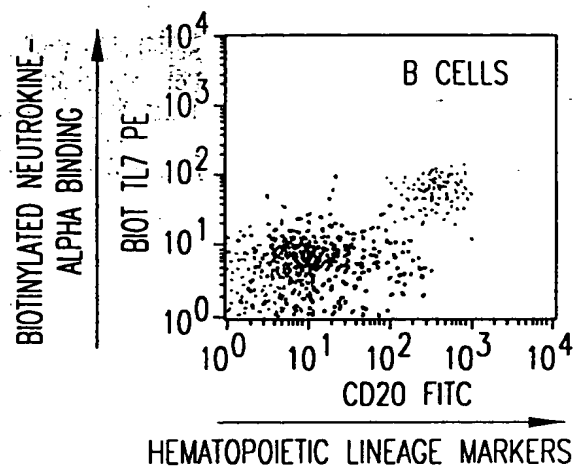


FIG.10B

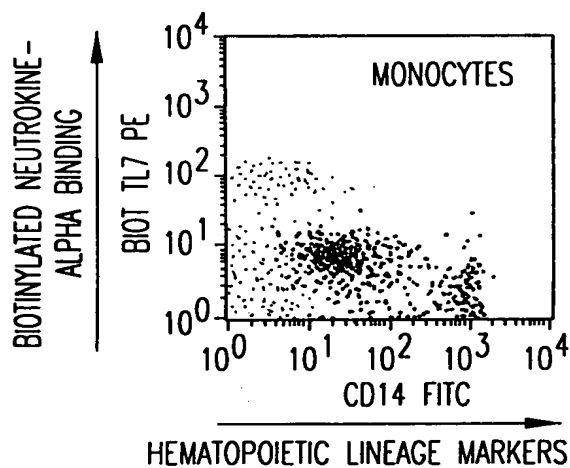


FIG.10C

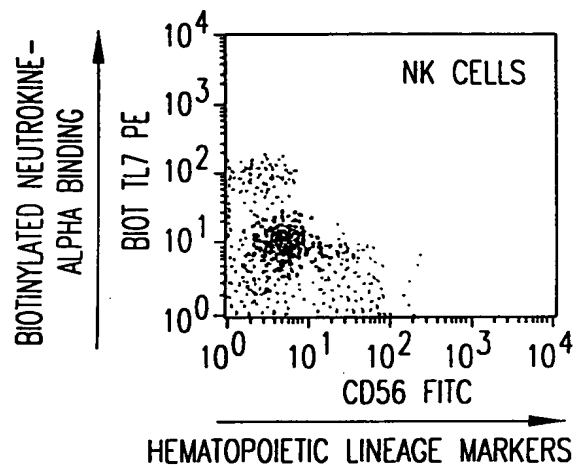


FIG.10D

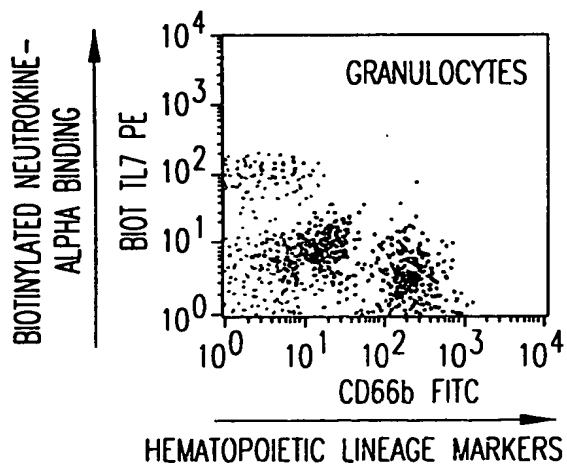


FIG.10E

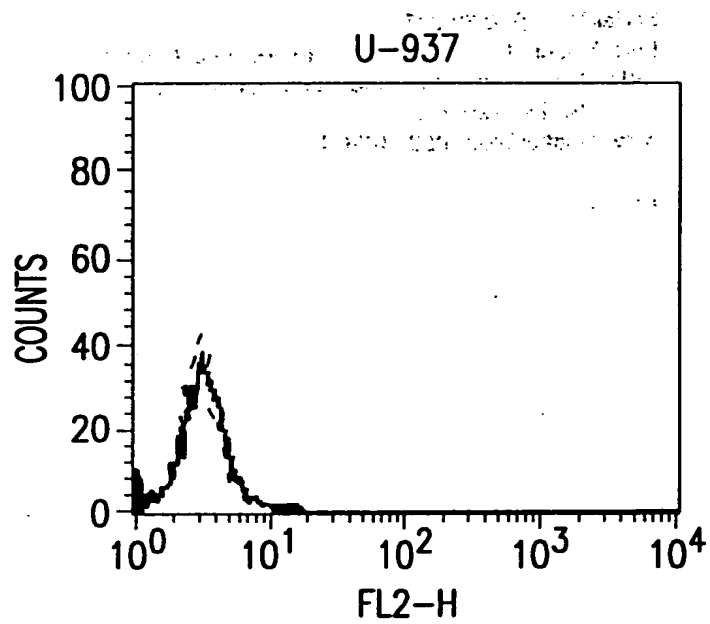


FIG.10F

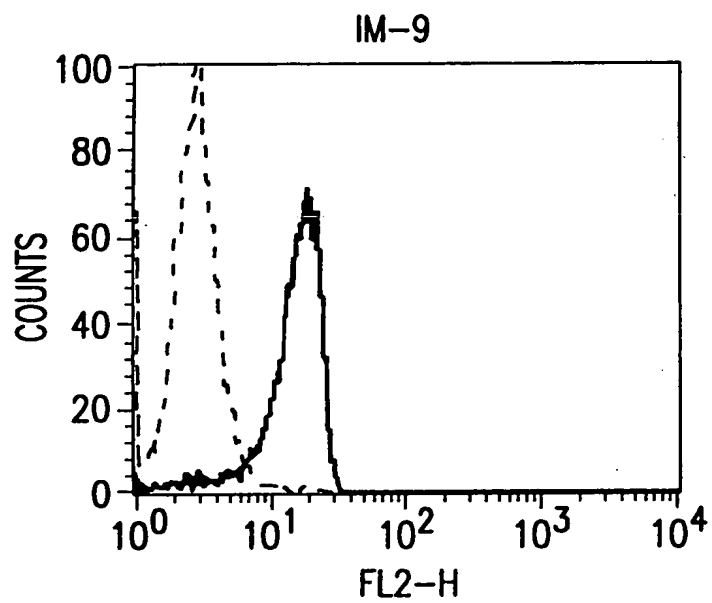


FIG.10G

Neutrokine - alpha
treated spleen
(2mg/Kg) bid 4d

Normal spleen



H & E (100X)

CD45R(B220)
(40X)



FIG.11A

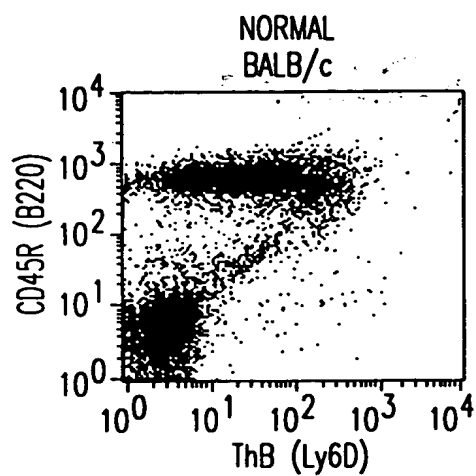


FIG. 11B

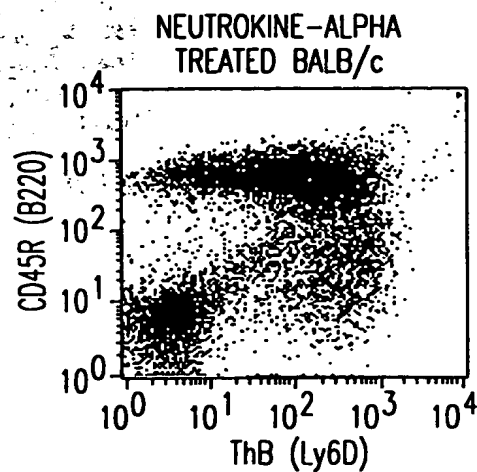


FIG. 11C

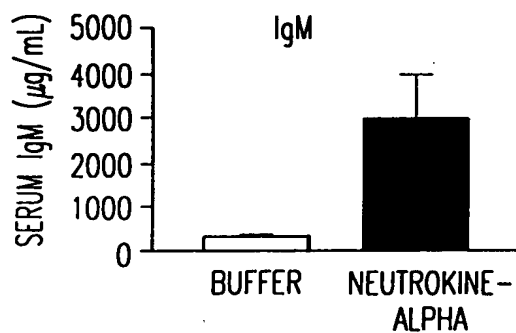


FIG. 11D

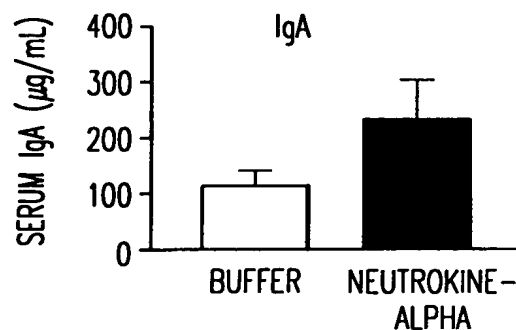


FIG. 11E

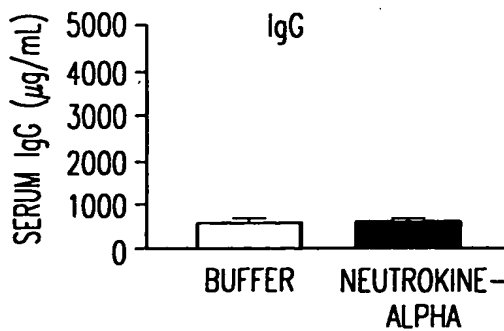


FIG. 11F